

## **REMARKS**

Favorable reconsideration of this application in light of the preceding amendments and the following remarks is respectfully requested.

Claims 10-13 having been added and no claims having been cancelled, the Applicants respectfully submit that claims 1-13 are pending with claims 3-13 remaining under consideration in this application, claims 1 and 2 having been withdrawn from consideration in response to the Applicant's previous election. Claims 1 and 3 are written in independent form.

The Applicants note with appreciation the Examiner's acknowledgement that certified copies of all priority documents have been received by the USPTO. Action Summary at 12.

The Applicants note with appreciation the Examiner's acknowledgement that the drawings filed with this application have been accepted by the Examiner. Action Summary at 11.

### **Elections/Restrictions**

The Applicant notes with appreciation the Examiner's acknowledgment of the Applicant's election of the Group II claims, claims 3-9, for prosecution in this application. Action at 2.

### **Objections to the Specification**

The Specification stands objected to as not being consecutively numbered. The Applicant hereby submits a substitute specification in which page numbering has been added. The Applicant submits that the addition of page numbers to the specification is sufficient to overcome this objection and request that it be reconsidered and removed accordingly.

### **Objections to the Drawings**

The Drawings stand objected to as including color photographs in the absence of a petition under 37 C.F.R. § 1.84(a)(2). The Applicant submits that this oversight is addressed by the petition filed herewith and requests that this objection be withdrawn upon the grant of the accompanying petition.

### **Claim Objections**

Claims 3, 5 and 6 stand objected to for various informalities as detailed in the Action. Action at 3-4. The Applicant submits that the amendments to the claims reflected above are sufficient to address and overcome each of the identified informalities. With respect to claim 4, the Applicant submits that the representations of the plasmids should be considered and treated as chemical formula in that they express, to those skilled in the art, the structure and relative positioning of the various chemical components comprising the plasmids. The Applicant requests, therefore, that these objections be withdrawn accordingly.

**Rejections under 35 U.S.C. § 112**

Claim 4 stands rejected under 35 U.S.C. § 112, first paragraph, as not being enabled by the original disclosure. The Applicant traverses this rejection for the reasons detailed below.

The Applicant submits that, as noted in paragraph [0024]:

A commercially available plasmid construct, pDsRed2-1 and p- $\alpha$ EGFPITR were selected as the basic materials. The plasmid construct pDsRed2-1 could be purchased from Clontech. The plasmid construct p- $\alpha$ EGFPITR could be produced according to the description of the related literature such as “Uniform GFP-expression in transgenic medaka (*Oryzias latipes*) at the F0 generation,” Chi-Yuan Chou et al., Transgenic Research 10: 303-315, 2001.

Accordingly, the Applicant submits that the materials and/or the techniques necessary to produce the recited plasmid are readily available to the public, and certainly to those of ordinary skill in the art, through commercial purchase from Clontech Corp. and/or from publications that would be known to those of ordinary skill in the art. The Applicant contends that one skilled in the art, guided by the present disclosure including, for example, paragraph [0024], and using only readily available materials would have no undue difficulty practicing the invention.

Accordingly, the Applicant submits that no deposit of the claimed plasmids is required under 37 C.F.R. § 1.802.

The Applicant submits that the amendments reflected above are sufficient to address the incubation period necessary for the successfully inoculated fertilized eggs to

express the fluorescent skeletal muscle and clarify that the skeletal muscle fluoresces rather than all tissues present in the transgenic golden zebrafish.

The Applicant requests, therefore, that this rejection be reconsidered and withdrawn accordingly.

### **Rejections under 35 U.S.C. § 103**

Claims 3, 4, 6, 7 and 9 stand rejected under 35 U.S.C. § 103(a) as unpatentable over Hsiao et al. [Developmental Dynamics, 220:323-36 (April 2001)] (“Hsiao”) in view of Carvan et al. [Ann. N.Y. Acad. Sci. 919:133-47 (2000)] (“Carvan”). Claims 5 and 8 stand rejected as unpatentable over Hsiao in view of Carvan and further in view of Finley et al. [Biotechniques, 31:66-72 (July 2001)]. The Applicant respectfully traverses these rejections for the reasons detailed below.

The Applicant contends that the proposed combinations of references are not sufficient to teach or suggest to a person having ordinary knowledge in the field the use of the red fluorescent protein gene to produce red fluorescent zebrafish according to the invention. The Applicant notes that Finley is directed to (i) 3-color imaging using fluorescent proteins in living zebrafish *embryos* and (ii) achieved expression of fluorescent genes in *mosaic form* as noted Finley’s Figure 3 and at page 68, column 2, line 23 and column 3, lines 1-4. The Applicant also notes that Finley teaches that DsRed expresses an orange-pink signal, Finley, page 68, column 2, lines 1-3.

The Applicant maintains that one of ordinary skill combining the Hsiao, Carvan and Finley references in the manner suggested would be led to an understanding that

certain fluorescent genes can be artificially expressed in zebrafish *embryos*, but contend that this focus on zebrafish embryos was necessitated by the presence of scales on the body of the adult zebrafish that would tend to block the desired fluorescence in the adult specimens. Accordingly, the Applicant suggests that these prior art references teach away from attempting to express such fluorescence, particularly red fluorescence, in adult zebrafish.

Further, the Applicant notes that invention relates to a method of producing a red fluorescent fish in which the red fluorescent is expressed throughout the whole body, *i.e.*, all skeletal muscle and not in mosaic form as achieved by the prior art methods. The Applicant submits that producing such a red fluorescent fish involves consideration of factors including, for example, the gene fragment construction, compatibility of species of the gene resource and the host, peptide folding, expression stability or protein turnover rate, whether the protein is expressed throughout the host body and whether the expression is readily visible in adult fish.

The Applicant further contends the efforts leading to the invention demonstrate that simply substituting a red fluorescent protein for the EGFP disclosed by Hsiao to achieve a viable red fluorescent adult zebrafish cannot be accomplished with a reasonable expectation of success. The Applicant spent many years trying various gene fragment constructions without success before identifying DsRed as a suitable gene for producing red fluorescent fish. Continued efforts have not yet identified another RFP that provides the results achieved with DsRed. Indeed, other apparently suitable genes resulted in either high mortality among the transgenic embryos, interfered with the maturation of the

transgenic embryos in some manner that prevented the development and/or survival of mature adult fish.

The Applicant maintains, therefore, that while certain of the components have been identified, the applied references are not sufficient, singly or in combination, to lead one of ordinary skill to the invention. The Applicant further suggests that the noted difficulties, and the non-obvious nature of the invention, are further demonstrated by the absence of commercially available red fluorescent fish despite a growing interest in such transgenic aquarium species as evidenced by TIME Magazine's selection of the Applicant's fluorescent fish as one of the "Coolest Inventions 2003" as reflected at (<http://www.time.com/time/2003/inventions/invfish.html>).

The Applicant maintains, therefore, that this invention was and remains a significant and non-obvious advancement in the art and requests, therefore, that these rejections be reconsidered and withdrawn.

### **CONCLUSION**


In view of the above remarks and amendments, the Applicants respectfully submit that each of the pending objections and rejections have been addressed and overcome, leaving the present application in condition for allowance. A notice to that effect is respectfully requested.

If the Examiner believes that personal communication will expedite prosecution of this application, the Examiner is invited to contact the undersigned.

If necessary, the Commissioner is hereby authorized in this, concurrent, and future replies to charge any underpayment or non-payment of any fees required under 37 C.F.R. §§ 1.16 or 1.17, or credit any overpayment of such fees, to Deposit Account No. 08-0750, including, in particular, extension of time fees.

Respectfully submitted,

HARNESS, DICKEY & PIERCE, P.L.C.

By:  (41,646) fm  
John A. Castellano  
Reg. No. 35,094

P.O. Box 8910  
Reston, VA 20195  
(703) 668-8000

JAC/GPB